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TOPICAL REVIEW

Form and function remixed: developmental physiology in the evolution of vertebrate body plans

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Abstract The most widely accepted model of evolutionary change, the Modern Evolutionary Synthesis, is based on the gradualism of Darwin and Wallace. They, in turn, developed their ideas in the context of 19th century concepts of how matter, including the tissues of animals and plants, could be reshaped and repatterned. A new physics of condensed, chemically, electrically and mechanically excitable materials formulated in the 20th century was, however, readily taken up by physiologists, who applied it to the understanding of dynamical, external condition-dependent and homeostatic properties of individual organisms. Nerve conduction, vascular and airway dynamics, and propagation of electrical excitations in heart and brain tissue all benefited from theories of biochemical oscillation, fluid dynamics, reaction–diffusion-based pattern instability and allied dissipative processes. When, in the late 20th century, the development of body and organ form was increasingly seen to involve dynamical, frequently non-linear processes similar to those that had become standard in physiology, a strong challenge to the evolutionary synthesis emerged. In particular, large-scale changes in organismal form now had a scientific basis other than gradualistic natural selection based on adaptive advantage. Moreover, heritable morphological changes were seen to be capable of occurring abruptly with little or no genetic change, with involvement of the external environment, and in preferred directions. This paper discusses three examples of morphological motifs of vertebrate bodies and organs, the somites, the skeletons of the paired limbs, and musculoskeletal novelties distinctive to birds, for which evolutionary origination and transformation can be understood on the basis of the physiological and biophysical determinants of their development.

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Introduction

Form and function are usually considered complementary but distinct aspects of living systems. The conviction that they require different modes of explanation is affirmed in the traditional disciplinary domains of anatomy and physiology. In the common conception, anatomy is like sculpture and physiology like music: the first is the outcome in space of the moulding of obdurate materials (flesh and bone), whereas the second is the playing out in time of transient or recurrent effects (e.g. glycaemic load, sleep and wakefulness).

The division between anatomy and physiology was reinforced by more than 150 years of adaptationist

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evolutionary theory. The early evolutionists were mostly morphologists. They were captivated by the fossil record and the impressive structural elaborations that distinguished animals from plants, and subtypes in those categories from one another. The reference standard of theoretical sufficiency in evolutionary theory was the ability to explain alterations in size and shape in genealogically related organisms. Given the poor understanding of the physics of condensed materials at the time when Charles Darwin and Alfred Russel Wallace were advancing their ideas, it is little wonder that they adopted a conservative, gradualistic notion of how forms change over time (Newman & Linde-Medina, 2013). The theory of evolution by advantage-driven natural selection in fact resembled the trial-and-error industrial practices of 19th century manufacturers like Darwin's maternal grandfather Josiah Wedgwood, who arrived at the breakthrough ceramic recipe and blue colouring of his most successful products only after many hundreds of trials (Dolan, 2004).

The classic theories of the evolution of form were thus committed to a model (natural selection of 'imperceptible variations') that considered only gradual modification; all other change (e.g. 'macroevolutionary' differences between phyla) was assumed to arise from many cycles of minuscule changes over very long periods of time. These models allowed no latitude for 'saltations' or sudden jumps between disparate forms that could originate new biological types. The possibility of these would have undermined the role of natural selection as the primary mode of morphological evolution.

Whereas some of his predecessors (e.g. Geoffroy Saint-Hilaire) were sympathetic to a role for the abrupt reorganization of body forms (Amundson, 2005), Darwin explicitly disavowed a role for the biological outliers that breeders of the time termed 'sports' (Darwin, 1868). This rejection in principle of rapid innovation in the anatomical realm, with some theoretical support from population genetics (Fisher, 1930), was carried over in the Modern Evolutionary Synthesis, the post-genetics revolution version of the adaptationist model.

However, living systems are dynamical entities and no Synthesis could prohibit physiologists from attending to abrupt changes in functional state, the most obvious one being death. Other physiological activities are similarly all-or-none in nature, and virtually all, including heart rate, emotional state and sexual readiness, are plastic and responsive to external conditions. Such phenotypic characters are troublesome for standard evolutionary theory because they depend on the environment as well as the genes, are often (as noted) abruptly divergent in their expression ('fight or flight'), and are typically buffered against perturbations ('homeostasis'). Notwithstanding this, physiology was not perceived as a threat to the standard evolutionary narrative because the abrupt transitions and conditionalities of physiological phenotypes could be represented (however problematically) as 'norms of reaction' of the genotype, with no irreversible impact on the next generation (Ostrowski *et al.* 2002). This idealization is often misleading, however, and is challenged by many recent studies (Jablonka & Raz, 2009; Bonduriansky & Day, 2013; Noble, 2013; Noble *et al.* 2013; Dias & Ressler, 2014).

Even so, inheritance of acquired physiological states is not usually associated with disparate morphologies, leaving the gradualism of the Synthesis relatively unchallenged on its traditional turf of anatomy for many decades. This changed late in the 20th century with the incorporation into evolutionary theory of embryology, which marked the rise of evolutionary developmental biology, or 'evo–devo' (Müller, 2007). Among the scientific strands that led to this new programme was a return to the idea that development is inherently physiological. Elucidation of feedback and feed-forward regulation in gene regulatory networks and reciprocity in cell–cell and cell–microenvironment interactions made this inevitable. In particular, the misleading 'genetic program' concept of embryonic development, advanced in the 1950s in analogy with digital computation [the principles of which emerged more or less simultaneously with those of molecular genetics (Cobb, 2013)], in which algorithmic instructions for ontogeny were sought in the organism's DNA, became increasingly untenable (Nijhout, 1990; Goodwin, 1994).

Soon, concepts from the new physics of non-linear dynamical systems and 'excitable media' (Winfree *et al.* 1985; Holden, 1990), which had provided a natural framework for understanding the physiology of nerve conduction and neurodynamics, renal countercurrent flow, cardiovascular and respiratory function, circadian rhythms, and so forth, were taken up by some developmental biologists (Gierer & Meinhardt, 1972; Newman & Frisch, 1979; Sager, 1996). These ideas were joined to results from the emerging field of 'soft matter' physics (de Gennes, 1992), which refers to viscoelastic matter acting on multiple scales (such as living tissues) to provide experimentally testable causal links (often in the form of mathematical and computational models) between genotypes and morphological phenotypes during embryogenesis.

If physics plays a causal role, along with genes, in the development of organismal form, there must be aspects of development and its outcomes (it was argued) that are 'generic' and are explicable in terms of processes that living systems have in common with non-living ones (Turing, 1952; Steinberg, 1978; Meinhardt, 1982; Newman & Comper, 1990). To be sure, the non-living physical systems invoked as models for developmental systems must have features not usually found together outside the living world. Animal embryos and regenerating tissues and organs consist of viscoelastic materials that are simultaneously chemically or mechanically excitable: they are capable of storing energy and thus responding to disturbances in an active rather than a passive fashion. The physics-based generic properties seen in developing systems could include such morphological motifs as tissue multilayering, and the formation of interior cavities, segments and appendages (Newman & Müller, 2000).

This 'physicalist' (or, in more contemporary terms, 'physico-genetic') viewpoint suggested, in turn, that ancient cell aggregates could have generated complex multicellular forms rapidly because the relevant physics was present from the start (Newman, 1994; Newman *et al.* 2006). This idea could resolve certain enigmas of the fossil record (Conway Morris, 2006) and gained support when, during the early part of the present century, it was recognized that the major portion of the gene set that mediates the morphological development of all animal types, the developmental–genetic 'toolkit', had been carried over from single-celled ancestors in which genes had evolved to perform unicellular rather than multicellular functions (Carroll *et al.* 2004). If novel genes could not account for the novel forms of the early metazoan radiations, additional causal agency was required (Newman, 2006). The typically non-linear physics of excitable, self-organizing soft matter, which continues to function in present-day developmental systems (Forgacs & Newman, 2005), appears to fulfil this requirement.

The idea that the origin of developmental systems was based on the dynamicity of multicellular clusters implies that subsequent selection for preservation of their morphological outcomes would have produced subsystem integration, physiological homeostasis, and the stabilization of developmental pathways referred to by Waddington (1957) as 'homeorhesis'. However, it is also inevitable that the developmentally plastic and environmentally conditioned aspects of such systems would have been retained to varying extents (Newman & Müller, 2000). In the following section, I will provide three examples of this plasticity–stability duality of 'developmental physiology' in the ontogeny and inferred phylogeny of morphological motifs of vertebrate embryos. These comprise: (i) a body plan motif common to all the taxonomic classes of vertebrates (segmentation); (ii) an organ-specific structural motif that differs markedly in form among and within the vertebrate classes (the tetrapod limb skeleton), and (iii) a suite of musculoskeletal modifications that sets one of the classes apart from the others (the avian body plan).

Dynamical innovation of vertebrate form: three examples

Clocks and gradients in vertebrate segmentation. The theory of non-linear oscillations, the mathematical roots of which derive from Poincaré's studies of dynamical systems in the late 19th century (Gray, 2012) and practical contributions to the field of electrical engineering in the 1920s (van der Pol, 1927), was taken up avidly by physiologists over the following decades. The Hodgkin–Huxley model of nerve conduction predicted both propagation and periodic changes in cellular electrical activity using a common, empirically based set of differential equations operating under different parameter choices (Hodgkin & Huxley, 1952). More generally, theorists and experimentalists recognized that oscillations of regulatory molecules and mechanical activity were enabled by the appropriate balance of positive and negative feedback interactions within individual cells and, with synchronization, globally in tissue masses (Goodwin, 1963; Marco & Nastuk, 1968). Increased understanding of the properties of these systems (Minorsky, 1962; Winfree, 1980; Goldbeter, 1996) led them to be applied to issues as varied as circadian rhythms (Winfree, 1974), the cardiac pacemaker (Guevara *et al.* 1981) and patterns of neural excitation in the brain (Traub & Bibbig, 2000).

Although systems that produce alternating states of activity or composition would seem naturally suited to account for the generation of serially repeated structures during embryogenesis, as well as the evolutionary innovation of such motifs, proposals along these lines were met with resistance. In the late 19th century William Bateson, an early geneticist, proposed a 'vibratory theory' of segmentation, in which he likened the process to the rippling of windblown sand, or the 'Chladni patterns' formed by powder placed on the back of an overturned violin. Bateson hoped to understand the action of genes by the physics of the systems in which they operate (Bateson & Bateson, 1928). This notion was treated dismissively by one key architect of the Synthesis (Mayr, 1982), but in the late 1990s experimental evidence demonstrated that vertebrate body segmentation (somitogenesis) was indeed generated by an intracellular biochemical oscillation. This was based in part on the feedback regulation of the transcriptional switching factor, Hes1, a downstream mediator of the Notch signalling pathway, in conjunction with an inhibitory gradient containing the morphogen FGF8, the source of which is at the embryo's tail tip (Palmeirim *et al.* 1997).

The dynamics of this system are formally similar to a theoretical 'clock-and-wavefront' mechanism proposed earlier by the developmental biologist Jonathan Cooke and the mathematician Christopher Zeeman. According to this idea, cells in the presomitic tissue oscillate in a synchronized fashion and their periodically changing cell state (the clock) acts as a 'gate' for the action of a determinant of potentially changed cell behaviour that sweeps along the embryo's length (the wavefront) (Cooke & Zeeman, 1976). The newer version of the mechanism also involves the elongation by growth of the embryo, whereby the dimensions and pace of formation of the somites are functions of clock period, gradient steepness and tissue elongation rate. The clock-and-wavefront mechanism may be superimposed on and limited in its efficacy by an intrinsic propensity of presomitic mesoderm to self-organize into discrete, somite-like tissue masses (Dias *et al.* 2014).

Segmentation appeared several times in widely divergent phyla, some without common segmented ancestors (Bateson, 1894; Willmer, 1990; Brusca & Brusca, 2003). This morphological motif presents challenges to gradualist evolutionary narratives (Minelli & Fusco, 2004) because segments are discrete structures that are added or lost in an all-or-none fashion, and also because large changes in segment number can occur in evolutionary lineages with little sign of intermediate forms. Oscillation-based mechanisms such as those described above, which embody the inherent discreteness of periodic processes and the environment-sensitive plasticity of dynamical systems, can help make sense of these developmental and evolutionary transitions.

In vertebrates, the numbers of somites in fish, birds and mammals are in the dozens, generally ranging between the 31 seen in zebrafish and the 65 in mouse. Snakes, by contrast, possess between 130 and about 500 (Marx & Rabb, 1972; Vonk & Richardson, 2008). Although arguments have been advanced for how this atypical segmental phenotype may have conferred adaptive advantages to snakes and their ancestors (Houssaye *et al.* 2013), a more fundamental question concerns how the developmental dynamics of somite formation are capable of generating such a divergent morphology.

Gómez *et al.* (2008) showed that corn snake embryos generate somites by a clock-and-wavefront mechanism similar to that of fish, birds and mammals, although they elaborate 10 times as many as the zebrafish. As in the embryos of other vertebrates, each new somite in a developing corn snake buds off for every recurrence of a critical clock-value. The possibilities that the snake had so many somites because its oscillator ran faster than that of the other vertebrates, or that the total length of the presomitic tissue, measured in cell numbers, was greater in these animals than in other vertebrates were considered. In fact, neither of these hypotheses is true. Instead, the rate of body elongation in the snake is considerably slower than that in chickens, mice and zebrafish because of the snake's low cell generation rate. The lizard *Aspidoscelis uniparens*, which has 90 somites, exhibits a similarly slow cell generation rate along its axis, but also has a very slow clock, with a period of about 4 h (Gómez *et al.* 2008).

The reason that the final somite number for the lizard falls within the 'normal' range of that in vertebrates is that the two key processes slowed down in a proportionate fashion during their evolution. As snakes emerged later in evolution than lizards, the slowing down of the clock and cell cycle may have occurred in the common squamate ancestor of these two groups. However, in the lineage that gave rise to snakes, a body plan with an exorbitant number of somites arose, presumably, with the restoration of the oscillation to a more typical vertebrate period range (Bhat & Newman, 2009).

Although one might envision a series of genetic changes affecting the characteristic rates of the regulatory processes underlying the oscillator, the fact that the segmentation clock is a dynamical biochemical system suggests that somitogenesis might also exhibit environment-dependent plasticity. Indeed, egg incubation temperature is known to affect vertebrae number in fish and snakes (Fowler, 1970; Osgood, 1978), and variations in the uterine environment have similar consequences in mammals (McLaren & Michie, 1958). Such physiological determination of the development of form is likely to have been even more pronounced when these body plan motifs first arose, before millions of years of 'canalizing' (Waddington, 1942) and stabilizing (Schmalhausen, 1949) evolution rendered species-specific morphological phenotypes relatively resistant to external effects (Newman, 2005).

The recognition that dynamical processes like the clock-and-wavefront mechanism underlie the generation of specific morphological motifs in present-day embryos makes the question of evolutionary *origination* of such constructional elements much easier to conceptualize than does the neo-Darwinian model. The supposition of the Modern Synthesis is that a complex biological structure like a segmented body plan must have arisen gradually and that marginally superior adaptation must have been the primary criterion for the prevalence of forms increasingly like the present-day ones. By contrast, tissues that are organized by physiological dynamics can undergo abrupt changes in pattern. In particular, oscillations and gradients could have existed in the embryos of unsegmented vertebrate ancestors for millions of years before a novel balance of rates resulting from mutational change, or a combination of mutation and environmental change, led to the relatively sudden and fortuitous appearance of a segmented body (Newman, 1993; Salazar-Ciudad *et al.* 2001*a*, *b*).

Dissipative structures in the vertebrate limb. In a paper titled 'The chemical basis of morphogenesis', the mathematician Alan Turing (1952) showed that a balance of positive and negative feedback in an open chemical system, when coupled with differences in the rates of diffusion of the key reactive molecules, could lead to a counterintuitive result. In defiance of the expectation that diffusion evens things out, such 'reaction–diffusion' systems were found mathematically to be capable of organizing into stable, non-uniform concentration patterns, often exhibiting periodicities. ['Reaction–diffusion' is used as shorthand when biological systems are concerned. Neither 'reaction' (i.e. cell-based production of, and response to, molecular factors) nor 'diffusion' (more generally, transport of released molecules through and between cells, often with the latter's active participation) are mechanistically like those seen in purely chemical systems.]

Although Turing had some unacknowledged predecessors in the analysis of such systems (Kolmogorov *et al.* 1937; Rashevsky, 1948), his lucid exposition soon began to have an impact in the fields of theoretical chemistry and biology. Ilya Prigogine, who would later win a Nobel Prize for extending these ideas, demonstrated a relationship between non-equilibrium thermodynamics and 'Turing patterns', coining the term 'dissipative structures' (Prigogine, 1969; Martinez, 1972). This framing emphasized the requirement for energy expenditure in maintaining the non-uniformities, situating the phenomenon at the interface of physiology and morphogenesis. By the early 1990s such patterns were unequivocally shown to occur in appropriately devised chemical systems (Castets *et al.* 1990; Ouyang & Swinney, 1991).

Within a few decades, physiologists studying perception adopted the reaction–diffusion formalism to model excitations in the visual cortex (Grossberg, 1976), for which it continues to be useful (Zucker, 2012). A similar situation pertains to attempts to understand the spatiotemporal dynamics of the normal and compromised myocardium (Winfree, 1980; Shiferaw & Karma, 2006; Potse *et al.* 2007).

The development of the quasi-periodically arranged skeletal elements of the vertebrate limb would seem to lend itself to explanations in terms of a Turing-type mechanism. However, the most widely discussed model for this phenomenon in the 1970s relied heavily on the popular cybernetic information paradigm of the period (Summerbell *et al.* 1973; Tickle *et al.* 1975). Specifically, the skeletal pattern was proposed to form by the implementation of a species-characteristic map between the values of a graded spatial or temporal signal ('positional information') and a hypothesized representation of the developing limb in the organism's genome (Wolpert, 1969).

An alternative mechanism based on the experimentally determined capacity of the limb's mesenchymal tissue to exhibit self-organizational properties that resembled those of Turing's chemical reaction–diffusion system was proposed at the end of the decade (Newman & Frisch, 1979). Little was known at the time about the cell and molecular interactions responsible for the mesenchymal condensations that provide the template for the bones, but the model used mathematical arguments to show that a succession of condensation patterns exhibiting proximodistally increasing numbers of parallel elements could emerge, in principle, under realistic changes in the size and shape of the undifferentiated tip of the limb bud. As more detailed biological information about this system emerged over the next decades, the model was sustained (Hentschel *et al.* 2004; Zhu *et al.* 2010; Glimm *et al.* 2014).

In addition to providing an explanation for why vertebrate limb skeletons have the quasi-periodic architecture they do [the positional information mechanism being specifically advertised as being unconstrained in the patterns it could generate, given a suitable genome (Wolpert, 1969)], Turing-type models (like the clock-and-wavefront mechanism for somitogenesis) exhibit the inherent plasticity of dynamical (as opposed to programmed) process. Models of this type have proved particularly suitable for accounting for the effects of limb-affecting mutations, such as the 'mixed-mode' (thick-thin) digit morphology of *Doublefoot* mouse mutants (Miura *et al.* 2006), the thinner, more densely packed digits seen in mouse embryos with progressive loss of distally expressed Hox genes (*Hoxa13* and *Hoxd11–13*) against a Gli3-null background (Sheth *et al.* 2012), and the spatially biased and discontinuous distribution of supernumerary digits in Maine Coon cats with point mutations in a cis-regulatory element of sonic hedgehog (Lange *et al.* 2014). Reaction–diffusion models of limb development have also provided insights (although necessarily speculative) into morphological variation seen in the fossil record of the tetrapods and their piscine antecedents (Zhu *et al.* 2010), as well as into the shift in digit identity that appears to have occurred during the evolution of birds from their presumed theropod dinosaur ancestors (Čapek *et al.* 2014).

As with the suggestion above concerning the relatively sudden origination of the segmented body plan of vertebrates, one can hypothesize that a re-tuning of parameters governing activatory and inhibitory mechanisms of cartilage development led to the novel formation of, and abrupt transitions between, quasi-periodic arrays of skeletal elements in what, in ancestral forms, were fin-like fleshy lobes (Zhu *et al.* 2010).

Thermogenesis in the origination of the avian body plan. After the emergence of the vertebrates (animals with segmented backbones), the tetrapods (vertebrates with paired limbs) and the amniotes (tetrapods that develop in an intra-egg sac), the possibility of further body plan innovation was still not exhausted. Bird skeletal anatomy differs markedly from that of the other amniotes (reptiles and mammals) (Kaiser, 2007). The bipedality of birds, for example, is distinct from that inferred for any of their dinosaur ancestors, being associated with unique skeletal modifications to the pelvis and the bones of the lower limb. Specifically, birds have no separate lumbar vertebrae, but rather a 'synsacrum', consisting of a variable (depending on the species) group of lower vertebrae fused to one another and to the pelvis. The synsacrum is independently mobile from the thoracic portion of the vertebral column, leading, with the associated interruption in the overlying muscles, to relatively autonomous anterior and posterior 'locomotor modules' in the avian body (Gatesy & Dial, 1996).

Several additional bird-specific modifications of the pelvis and legs are associated with functional adaptations of this vertebrate class, such as unfused pubic bones that permit the passage of eggs that are large relative to body size, plates of an expanded pelvic ischium that protect a digestive system more elaborate that that of other reptiles, and unique specializations of the avian leg in which skeletal novelties derived from the fusion of ancestral bones, the tibiotarsus, syndesmosis tibiofibularis and tarsometatarsus, facilitate locomotory patterns of walking, running and swimming unlike those of other tetrapods. The anterior locomotor module of birds contains its own skeletal novelties in addition to the highly disparate forelimbs, which became vestigial in some dinosaurs and birds, and evolved into wings and paddles in other birds. These include the keel, an extension of the sternum that provides an anchor for the breast muscles, and the furcula (wishbone), formed by the fusion of the clavicles (Kaiser, 2007).

Standard (i.e. Modern Synthesis-based) evolutionary scenarios would suggest that these skeletal innovations arose through successive cycles of natural selection in an ancestral saurian lineage, with anatomical architectures suitable for flying, swimming or bipedal locomotion emerging after millions of years. However, the plasticity of embryonic skeletogenesis provides an alternative basis by which they may have emerged in a concerted fashion, without the requirement for incremental adaptive advantage over successive generations. In particular, several lines of evidence indicate that the skeletal muscle-dependent mechanical activity of the embryo is required for the normal ontogeny of skeletal elements of vertebrates, particularly in birds (Nowlan *et al.* 2010; Newman, 2011; Newman *et al.* 2013). Paralysed chick embryos, for example, fail to form the fibular crest, which was a morphological novelty in theropod dinosaurs and is a necessary component in the development of the avian-specific tibiofibularis (Müller & Streicher, 1989).

In addition, those portions of the thoracic skeleton that most distinguish birds from other tetrapods are also those most dependent on muscular activity for their normal morphogenesis. The clavicles, for example, which are fused into the furcula in birds, were underdeveloped and unfused in chick embryos in which muscles were paralysed (Hall & Herring, 1990; Hall, 2001). Given the responsiveness of the developing avian skeleton to mechanical forces, it is plausible that many bird-specific skeletal innovations reflect the presence of large skeletal muscles in this group, particularly in comparison with related taxa such as lizards or mammals.

Depending on whether a bird is flightless or volant, a runner or swimmer, it will have either hyperplastic thigh muscles, breast muscles, or both. Despite the broad morphological diversity of the birds, the presence of large depots of skeletal muscle in either or both the anterior and posterior body regions is a unifying motif across all the specialized avian orders. Was the evolution of these massive muscles a coordinated response to the same selective forces that led to the reshaping of the avian skeleton? If so, why did it occur independently, and to very different functional effects, in the anterior and posterior locomotor modules? Here, whole-organism physiology, in conjunction with phylogenomics, provides an alternative solution to this problem, in which skeletal muscles were under directional selective pressure, driven by the internal requirements of the organism, to increase in size. Under this scenario, the trend toward increasingly larger skeletal muscles in the early stages of bird evolution was the key to the distinctive innovations [some of them developmental side-effects (Müller, 1990)] of the avian body plan.

The reasoning is as follows: among the most obvious of the physiological transformations accompanying the evolutionary emergence of mammals, birds and non-avian reptiles from their common ancestor was the diversification into homeothermic ('warm-blooded': mammals, birds) and poikilothermic ('cold-blooded': non-avian reptiles) classes. Mammals branched off first. Among several specializations contributing to their body heat generation was the recruitment of an ancient, nuclear gene-encoded mitochondrial protein, uncoupling protein-1 (UCP1), into cold-induced physiological pathways in mammal-specific thermogenic brown and beige adipose tissues (Wu *et al.* 2012). UCP1 causes leakage of the inner mitochondrial membrane, generating heat by uncoupling oxidative phosphorylation. The gene *UCP1* is absent in birds and non-avian reptiles, including turtles (Mezentseva *et al.* 2008; Newman *et al.* 2013); it was presumably deleted in a sauropsid ancestor.

Because the common ancestor of sauropsids and mammals probably had an intact thermogenic fat programme (Mezentseva *et al.* 2008; Newman *et al.* 2013), it must have been endothermic, or at least heterothermic (Grigg *et al.* 2004). The loss of *UCP1* would have presented these animals with a crisis. Some descendents adopted a poikilothermic physiology, giving rise to modern non-avian reptiles. Birds, however, retained a capacity for endothermy and, ultimately, homeothermy. Avian skeletal muscles eventually evolved a number of biochemical and physiological specializations for heat generation (Newman *et al.* 2013). Initially, however, in the population of *UCP1*-lacking sauropsid ancestors which eventually produced the birds, possibly enlarged skeletal muscles and, more certainly, muscle hyperplasia (driven by selection for enhanced thermogenesis) over the course of subsequent evolution permitted these animals to thrive as endotherms in increasingly colder venues, albeit with unique muscle-laden anatomies.

The massive thigh and pectoral muscles would have forced the acquisition of bipedality in some groups and facilitated the capability for swimming and flight in others (Newman, 2011). Concomitant with its aforementioned role in generating an array of opportunistically recruited skeletal novelties, hyperplastic skeletal musculature enabled birds to overcome the genetic deficit incurred by the loss of *UCP1* in their ancestors to become the most species-rich and ecologically diverse class of land vertebrates.

Discussion

I have reviewed the involvement of dynamical, condition-dependent (i.e. physiological in the broad sense) mechanisms in the anatomical organization of the vertebrate body. My main goal was to examine the roles of these mechanisms and processes in the evolutionary origination and transformation of certain structural features of these body plans. However, as the morphological determinants of fossil forms are outside the realm of experimentation, and it is well-accepted that present-day vertebrates are descendents of organisms that bore the same or similar features hundreds of millions of years ago, it is inevitable that the origination question would be addressed from the perspective of the developmental biology of present-day organisms.

The developmental processes discussed pertain to multiple levels of structural organization in vertebrate bodies and draw on a variety of biophysical effects that up until recently have figured more prominently in the physiological than the anatomical sciences. Somitogenesis, a process common to all vertebrate organisms that is in fact responsible for their defining character, was seen to involve intracellular biochemical oscillations and their synchronization across multicellular domains. Such mechanisms are a perennial of physiology at a variety of functional and temporal scales (Weber, 2009; Arnal & Giraud, 2012). Limb skeleton formation and patterning make use of the spatial propagation of excitatory activity, a feature common to the Turing-type reaction–diffusion mechanism described, and to a vast number of physiological activities, from the action potential (Hodgkin & Huxley, 1952) to myocardial waves (Steinberg *et al.* 2006) and respiration (Clavica *et al.* 2009). Finally, the physiology of temperature regulation and, more particularly, its compromise in the *UCP1*-lacking lineage of amniotes that ultimately gave rise to birds were seen to exert an indirect but decisive effect on the unique body plan of the avian vertebrate class.

Although the use of physiological concepts in an evo–devo consideration of body plan origination seems uncontroversial, the conclusions of such analyses can conflict with standard notions of how evolution of form has occurred. As physiological mechanisms are generally responsive to external conditions, extending them beyond their traditional roles in the homeostasis and facultative functioning of the postnatal organism to embryonic development raises the spectre of environmental plasticity and the inheritance of acquired characteristics (West-Eberhard, 2003). Further, because they typically involve often non-linear dynamic processes, their incorporation into the genotype–phenotype ‘map’ for the generation of form must accommodate the possibility of the abrupt appearance of morphological novelties.

By the mechanisms described here, morphological novelties can potentially arise in the space of one or a few generations, particularly in forms that are not extensively canalized (e.g. evolutionarily ancient ones). This suggests that the paleontological scenario of ‘punctuated equilibrium’ (Eldredge & Gould, 1972) may appropriately be applied in the iconoclastic spirit in which it was originally proposed, as ‘an alternative to phyletic gradualism’, rather than in the populational, Modern Synthesis version ultimately adopted by Stephen Jay Gould (2002).

Lastly, the speculation that the loss of a uniquely important gene (a usually fatal occurrence in the neo-Darwinian narrative) could actually predispose a lineage to mobilize pre-existing physiological functions to eke out bare survival, and then undergo biochemical and morphological evolution driven by the need to compensate for a function deficit, invokes two taboos of the standard model: unidirectional evolution, the rationale for which is discussed above, and the survival of phenotypic outliers. Concerning the latter, once we acknowledge the existence of a set of heritable mechanisms for morphological change that operate independently of incremental adaptive advantage, the complementary Modern Synthesis notion of a prohibition against deviation can be strongly questioned. As has been long maintained by Richard Lewontin (1983) and eloquently asserted, with newer evidence, by Patrick Bateson (2014): ‘The environment does not simply set a problem to which the organism has to find a solution. The organism can do a great deal to create an environment to which it is best suited.’

In his famous monograph on the history of biological thought, Ernst Mayr (1982) asserted: ‘Nothing strengthened the theory of natural selection as much as the refutation, one by one, of all the competing theories, such as saltationism, orthogenesis, [and] inheritance of acquired characters.' I have argued that newer evidence from experimental embryology, interpreted in the light of concepts from condensed matter physics and physiology unknown at the time of Darwin and his early 20th century successors, challenges this tenet and each of its components. This challenge does not outright abolish gradualist natural selection, but it does relegate it to a role in the fine-tuning and refining of heritable modifications that arise by other, often physiologically based, means.

References

Amundson R (2005). *The Changing Role of the Embryo in Evolutionary Thought: Roots of Evo–Devo*. Cambridge University Press, Cambridge; New York, NY.

Arnal LH & Giraud AL (2012). Cortical oscillations and sensory predictions. *Trends Cogn Sci* **16**, 390–398.

Bateson P (2014). New thinking about biological evolution. *Biol J Linn Soc Lond* (in press; doi: 10.1111/bij.12125).

Bateson W (1894). *Materials for the Study of Sariation*. Macmillan, London.

Bateson W & Bateson B (1928). *William Bateson, F. R. S., Naturalist; His Essays & Addresses, Together with a Short Account of his Life*. University Press, Cambridge.

Bhat R & Newman SA (2009). Snakes and ladders: the ups and downs of animal segmentation. *J Biosci* **34**, 163–166.

Bonduriansky R & Day T (2013). Nongenetic inheritance and the evolution of costly female preference. *J Evol Biol* **26**, 76–87.

Brusca RC & Brusca GJ (2003). *Invertebrates*. Sinauer Associates, Sunderland, MA.

Čapek D, Metscher BD & Müller GB (2014). Thumbs down: a molecular-morphogenetic approach to avian digit homology. *J Exp Zoolog B Mol Dev Evol* **322**, 1–12.

Carroll SB, Grenier JK & Weatherbee SD (2004). *From DNA to Diversity: Molecular Genetics and the Evolution of Animal Design*. Blackwell Publishing, Malden, MA.

Castets V, Dulos E, Boissonade J & DeKepper P (1990). Experimental evidence of a sustained standing Turing-type nonequilibrium chemical pattern. *Phys Rev Lett* **64**, 2953–2956.

Clavica F, Alastruey J, Sherwin SJ & Khir AW (2009). One-dimensional modelling of pulse wave propagation in human airway bifurcations in space-time variables. *Conf Proc IEEE Eng Med Biol Soc* **2009**, 5482–5485.

Cobb M (2013). 1953: when genes became 'information'. *Cell* **153**, 503–506.

Conway Morris S (2006). Darwin's dilemma: the realities of the Cambrian 'explosion'. *Philos Trans R Soc Lond B Biol Sci* **361**, 1069–1083.

Cooke J & Zeeman EC (1976). A clock and wavefront model for control of the number of repeated structures during animal morphogenesis. *J Theor Biol* **58**, 455–476.

Darwin C (1868). *The Variation of Animals and Plants under Domestication*. J. Murray, London.

de Gennes PG (1992). Soft matter. *Science* **256**, 495–497.

Dias AS, de Almeida I, Belmonte JM, Glazier JA & Stern CD (2014). Somites without a clock. *Science* **343**, 791–795.

Dias BG & Ressler KJ (2014). Parental olfactory experience influences behavior and neural structure in subsequent generations. *Nat Neurosci* **17**, 89–96.

Dolan B (2004). *Wedgwood: the First Tycoon*. Viking, New York, NY.

Eldredge N & Gould SJ (1972). Punctuated equilibria: an alternative to phyletic gradualism. In *Models in Paleobiology*, ed. Schopf TJM, pp. 82–115. Freeman, Cooper & Co., San Francisco, CA.

Fisher RA (1930). *The Genetical Theory of Natural Selection*. Clarendon Press, Oxford.

Forgacs G & Newman SA (2005). *Biological Physics of the Developing Embryo*. Cambridge University Press, Cambridge.

Fowler JA (1970). Control of vertebral number in teleosts – an embryological problem. *Q Rev Biol* **45**, 148–164.

Gatesy S & Dial K (1996). Locomotor modules and the evolution of avian flight. *Evolution* **50**, 331–340.

Gierer A & Meinhardt H (1972). A theory of biological pattern formation. *Kybernetik* **12**, 30–39.

Glimm T, Bhat R & Newman SA (2014). Modeling the morphodynamic galectin patterning network of the developing avian limb skeleton. *J Theor Biol* **346**, 86–108.

Goldbeter A (1996). *Biochemical Oscillations and Cellular Rhythms: the Molecular Bases of Periodic and Chaotic Behaviour*. Cambridge University Press, Cambridge.

Gómez C, Ŏzbudak EM, Wunderlich J, Baumann D, Lewis J & Pourquié O (2008). Control of segment number in vertebrate embryos. *Nature* **454**, 335–339.

Goodwin BC (1963). *Temporal Organization in Cells; a Dynamic Theory of Cellular Control Processes*. Academic Press, London; New York, NY.

Goodwin BC (1994). *How the Leopard Changed its Spots*. Weidenfeld & Nicolson, London.

Gould SJ (2002). *The Structure of Evolutionary Theory*. Belknap Press of Harvard University Press, Cambridge, MA.

Gray J (2012). *Henri Poincaré: a Scientific Biography*. Princeton University Press, Princeton, NJ.

Grigg GC, Beard LA & Augee ML (2004). The evolution of endothermy and its diversity in mammals and birds. *Physiol Biochem Zool* **77**, 982–997.

Grossberg S (1976). On the development of feature detectors in the visual cortex with applications to learning and reaction–diffusion systems. *Biol Cybern* **21**, 145–159.

Guevara MR, Glass L & Shrier A (1981). Phase locking, period-doubling bifurcations, and irregular dynamics in periodically stimulated cardiac cells. *Science* **214**, 1350–1353.

Hall BK (2001). Development of the clavicles in birds and mammals. *J Exp Zool* **289**, 153–161.

Hall BK & Herring SW (1990). Paralysis and growth of the musculoskeletal system in the embryonic chick. *J Morphol* **206**, 45–56.

Hentschel HG, Glimm T, Glazier JA & Newman SA (2004). Dynamical mechanisms for skeletal pattern formation in the vertebrate limb. *Proc R Soc Lond B Biol Sci* **271**, 1713–1722.

Hodgkin AL & Huxley AF (1952). A quantitative description of membrane current and its application to conduction and excitation in nerve. *J Physiol* **117**, 500–544.

Holden AV (1990). Some mathematical problems of excitability. *Biomed Biochim Acta* **49**, 651–660.

Houssaye A, Boistel R, Bohme W & Herrel A (2013). Jack-of-all-trades master of all? Snake vertebrae have a generalist inner organization. *Naturwissenschaften* **100**, 997–1006.

Jablonka E & Raz G (2009). Transgenerational epigenetic inheritance: prevalence, mechanisms, and implications for the study of heredity and evolution. *Q Rev Biol* **84**, 131–176.

Kaiser GW (2007). *The Inner Bird: Anatomy and Evolution.* UBC Press, Vancouver, BC.

Kolmogorov A, Petrovsky L & Piskunov N (1937). An investigation of the diffusion equation combined with an increase in mass and its application to a biological problem. *Bull Uni Moscow Ser Int A1* **6**, 1–26.

Lange A, Nemeschkal HL & Müller GB (2014). Biased polyphenism in polydactylous cats carrying a single point mutation: the Hemingway model for digit novelty. *J Exp Zoolog B Mol Dev Evol* (in press).

Lewontin RC (1983). Gene, organism and environment. In *Evolution from Molecules to Men*, ed. Bendall D, pp. 273–285. Cambridge University Press, Cambridge.

Marco LA & Nastuk WL (1968). Sarcomeric oscillations in frog skeletal muscle fibers. *Science* **161**, 1357–1358.

Martinez HM (1972). Morphogenesis and chemical dissipative structures. A computer simulated case study. *J Theor Biol* **36**, 479–501.

Marx H & Rabb GB (1972). Phyletic analysis of fifty characters of advanced snakes. *Fieldiana Zool* **63**, 1–321.

Mayr E (1982). *The Growth of Biological Thought: Diversity, Evolution, and Inheritance*. Belknap Press, Cambridge, MA.

McLaren A & Michie D (1958). An effect of the uterine environment upon skeletal morphology in the mouse. *Nature* **181**, 1147–1148.

Meinhardt H (1982). *Models of Biological Pattern Formation.* Academic Press, New York, NY.

Mezentseva NV, Kumaratilake JS & Newman SA (2008). The brown adipocyte differentiation pathway in birds: an evolutionary road not taken. *BMC Biol* **6**, 17.

Minelli A & Fusco G (2004). Evo–devo perspectives on segmentation: model organisms, and beyond. *Trends Ecol Evol* **19**, 423–429.

Minorsky N (1962). *Nonlinear Oscillations*. Van Nostrand, Princeton, NJ.

Miura T, Shiota K, Morriss-Kay G & Maini PK (2006). Mixed-mode pattern in *Doublefoot* mutant mouse limb – Turing reaction–diffusion model on a growing domain during limb development. *J Theor Biol* **240**, 562–573.

Müller GB (1990). Developmental mechanisms at the origin of morphological novelty: a side-effect hypothesis. In *Evolutionary Innovations*, ed. Nitecki M, pp. 99–130. University of Chicago Press, Chicago, IL.

Müller GB (2007). Evo–devo: extending the evolutionary synthesis. *Nat Rev Genet* **8**, 943–949.

Müller GB & Streicher J (1989). Ontogeny of the syndesmosis tibiofibularis and the evolution of the bird hindlimb: a caenogenetic feature triggers phenotypic novelty. *Anat Embryol* **179**, 327–339.

Newman SA (1993). Is segmentation generic? *BioEssays* **15**, 277–283.

Newman SA (1994). Generic physical mechanisms of tissue morphogenesis: a common basis for development and evolution. *J Evol Biol* **7**, 467–488.

Newman SA (2005). The pre-Mendelian, pre-Darwinian world: shifting relations between genetic and epigenetic mechanisms in early multicellular evolution. *J Biosci* **30**, 75–85.

Newman SA (2006). The developmental–genetic toolkit and the molecular homology–analogy paradox. *Biol Theory* **1**, 12–16.

Newman SA (2011). Thermogenesis, muscle hyperplasia, and the origin of birds. *Bioessays* **33**, 653–656.

Newman SA & Comper WD (1990). 'Generic' physical mechanisms of morphogenesis and pattern formation. *Development* **110**, 1–18.

Newman SA, Forgacs G & Müller GB (2006). Before programs: the physical origination of multicellular forms. *Int J Dev Biol* **50**, 289–299.

Newman SA & Frisch HL (1979). Dynamics of skeletal pattern formation in developing chick limb. *Science* **205**, 662–668.

Newman SA & Linde-Medina M (2013). Physical determinants in the emergence and inheritance of multicellular form. *Biol Theory* **8**, 274–285.

Newman SA, Mezentseva NV & Badyaev AV (2013). Gene loss, thermogenesis, and the origin of birds. *Ann N Y Acad Sci* **1289**, 36–47.

Newman SA & Müller GB (2000). Epigenetic mechanisms of character origination. *J Exp Zoolog B Mol Dev Evol* **288**, 304–317.

Nijhout HF (1990). Metaphors and the roles of genes in development. *BioEssays* **12**, 441–446.

Noble D (2013). Physiology is rocking the foundations of evolutionary biology. *Exp Physiol* **98**, 1235–1243.

Noble DW, Wechmann K, Keogh JS & Whiting MJ (2013). Behavioral and morphological traits interact to promote the evolution of alternative reproductive tactics in a lizard. *Am Nat* **182**, 726–742.

Nowlan NC, Sharpe J, Roddy KA, Prendergast PJ & Murphy P (2010). Mechanobiology of embryonic skeletal development: insights from animal models. *Birth Defects Res C Embryo Today* **90**, 203–213.

Osgood DW (1978). Effects of temperature on the development of meristic characters in *Natrix fasciata. Copeia* **1**, 33–47.

Ostrowski MF, Jarne P, Berticat O & David P (2002). Ontogenetic reaction norm for binary traits: the timing of phallus development in the snail *Bulinus truncatus. Heredity (Edinb)* **88**, 342–348.

Ouyang Q & Swinney H (1991). Transition from a uniform state to hexagonal and striped Turing patterns. *Nature* **352**, 610–612.

Palmeirim I, Henrique D, Ish-Horowicz D & Pourquié O (1997). Avian hairy gene expression identifies a molecular clock linked to vertebrate segmentation and somitogenesis. *Cell* **91**, 639–648.

Potse M, Coronel R, Opthof T & Vinet A (2007). Simulating T-wave parameters of local extracellular electrograms with a whole-heart bidomain reaction–diffusion model: size matters! *Conf Proc IEEE Eng Med Biol Soc* **2007**, 6644–6647.

Prigogine I (1969). Structure, dissipation and life. In *Theoretical Physics and Biology*, ed. Marios M, pp. 23–52. North Holland Publishing, Amsterdam.

Rashevsky N (1948). *Mathematical Biophysics*. University of Chicago Press, Chicago, IL.

Sager BM (1996). Propagation of traveling waves in excitable media. *Genes Dev* **10**, 2237–2250.

Salazar-Ciudad I, Newman SA & Solé RV (2001*a*). Phenotypic and dynamical transitions in model genetic networks. I. Emergence of patterns and genotype–phenotype relationships. *Evol Dev* **3**, 84–94.

Salazar-Ciudad I, Solé RV & Newman SA (2001*b*). Phenotypic and dynamical transitions in model genetic networks. II. Application to the evolution of segmentation mechanisms. *Evol Dev* **3**, 95–103.

Schmalhausen II (1949). *Factors of Evolution*. Blakiston, Philadelphia, PA.

Sheth R, Marcon L, Bastida MF, Junco M, Quintana L, Dahn R, Kmita M, Sharpe J & Ros MA (2012). Hox genes regulate digit patterning by controlling the wavelength of a Turing-type mechanism. *Science* **338**, 1476–1480.

Shiferaw Y & Karma A (2006). Turing instability mediated by voltage and calcium diffusion in paced cardiac cells. *Proc Natl Acad Sci U S A* **103**, 5670–5675.

Steinberg BE, Glass L, Shrier A & Bub G (2006). The role of heterogeneities and intercellular coupling in wave propagation in cardiac tissue. *Philos Trans A Math Phys Eng Sci* **364**, 1299–1311.

Steinberg MS (1978). Specific cell ligands and the differential adhesion hypothesis: how do they fit together? In *Specificity of Embryological Interactions*, ed. Garrod DR, pp. 97–130. Chapman & Hall, London.

Summerbell D, Lewis JH & Wolpert L (1973). Positional information in chick limb morphogenesis. *Nature* **244**, 492–496.

Tickle C, Summerbell D & Wolpert L (1975). Positional signalling and specification of digits in chick limb morphogenesis. *Nature* **254**, 199–202.

Traub RD & Bibbig A (2000). A model of high-frequency ripples in the hippocampus based on synaptic coupling plus axon–axon gap junctions between pyramidal neurons. *J Neurosci* **20**, 2086–2093.

Turing AM (1952). The chemical basis of morphogenesis. *Philos Trans R Soc Lond B Biol Sci* **237**, 37–72.

van der Pol B (1927). On relaxation–oscillations. *Philos Mag J Sci* **2**, 978–992.

Vonk FJ & Richardson MK (2008). Developmental biology: serpent clocks tick faster. *Nature* **454**, 282–283.

Waddington CH (1942). Canalization of development and the inheritance of acquired characters. *Nature* **150**, 563–565.

Waddington CH (1957). *The Strategy of the Genes*. Allen & Unwin, London.

Weber F (2009). Remodeling the clock: coactivators and signal transduction in the circadian clockworks. *Naturwissenschaften* **96**, 321–337.

West-Eberhard MJ (2003). *Developmental Plasticity and Evolution*. Oxford University Press, Oxford; New York, NY.

Willmer P (1990). *Invertebrate Relationships*. Cambridge University Press, Cambridge.

Winfree AT (1974). Suppressing drosophila circadian rhythm with dim light. *Science* **183**, 970–972.

Winfree AT (1980). *The Geometry of Biological Time*. Springer-Verlag, New York, NY.

Winfree AT, Winfree EM & Seifert H (1985). Organizing centers in cellular excitable media. *Physica* **17D**, 109–115.

Wolpert L (1969). Positional information and the spatial pattern of cellular differentiation. *J Theoret Biol* **25**, 1–47.

Wu J, Bostrom P, Sparks LM, Ye L, Choi JH, Giang AH, Khandekar M, Virtanen KA, Nuutila P, Schaart G, Huang K, Tu H, van Marken Lichtenbelt WD, Hoeks J, Enerback S, Schrauwen P & Spiegelman BM (2012). Beige adipocytes are a distinct type of thermogenic fat cell in mouse and human. *Cell* **150**, 366–376.

Zhu J, Zhang YT, Alber MS & Newman SA (2010). Bare bones pattern formation: a core regulatory network in varying geometries reproduces major features of vertebrate limb development and evolution. *PLoS One* **5**, e10892.

Zucker SW (2012). Local field potentials and border ownership: a conjecture about computation in visual cortex. *J Physiol Paris* **106**, 297–315.

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